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Brief Communication

Highly Enantioselective Catalytic Synthesis of Neurite Growth-Promoting Secoyohimbanes

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SUMMARY

Natural products endowed with neuromodulatory activity and their underlying structural scaffolds may inspire the synthesis of novel neurotrophic compound classes. The spirocyclic secoyohimbane alkaloid rhynchophylline is the major component of the extracts of *Uncaria* species used in Chinese traditional medicine for treatment of disorders of the central nervous system. Based on the structure of rhynchophylline, a highly enantioselective and efficient organocatalyzed synthesis method was developed that gives access to the tetracyclic secoyohimbane scaffold, embodying a quaternary and three tertiary stereogenic centers in a one-pot multistep reaction sequence. Investigation of a collection of the secoyohimbanes in primary rat hippocampal neurons and embryonal stem cell-derived motor neurons led to discovery of compounds that promote neurite outgrowth and influence the complexity of neuronal network formation.

INTRODUCTION

Many neurological diseases are characterized by a progressive loss of neuronal activity, and novel approaches aimed at restoration of neuronal viability and complexity of neuronal networks or at prevention of neuronal decline are in high demand. Therefore, the identification of small molecules with neurotrophic or neuroprotective properties is of great current interest.

Frequently, natural products display this kind of activity (Wilson and Danishefsky, 2006; Carcache et al., 2006; Inoue et al., 2007; Rawat et al., 2008; Chen et al., 2009; Jessen et al., 2009; Schmidt et al., 2009; Jang et al., 2010; Jana et al., 2011; Jessen et al., 2011; Cheng et al., 2012; Mehta et al., 2012; Praveen Kumar et al., 2012), and their characteristic scaffolds define biologically prevalidated starting points in vast chemical structure space, inspiring the design and synthesis of compound collections endowed with the same or similar kind of biological activity (Li and Vederas, 2009; Newman and Cragg, 2007; Antonchick et al., 2010; Bon and Waldmann, 2010; Koch et al., 2005; Kumar and Waldmann, 2009; Nören-Müller et al., 2006, 2008; Waldmann et al., 2008).

With respect to neurotrophic and neuroprotective activity, an interesting case is the alkaloid rhynchophylline (1). Rhynchophylline is a major component of the extracts of *Uncaria* species, which are widely used in traditional Chinese medicine to treat ailments of the central nervous system. Alkaloids are the active pharmacological components in *Uncaria* species and make up about 0.2% of their dry weight. Rhynchophylline accounts for up to 50% of the total amount of alkaloids in these plants and exhibits similar pharmacological activity as the plant extract (Zhou and Zhou, 2010, 2012).

Rhynchophylline (1) and its isomer isorhynchophylline (2) embody the secoyohimbane scaffold (Figure 1; Shellard and Lala, 1978; Shellard et al., 1969; Phillipson and Hemingway, 1973a, 1973b). The key structural feature of these molecules is a complex spiro ring fusion at the three position of the oxindole core and the one position of an octahydroindolizine. These alkaloids often occur as pairs of interconvertible isomers (Seaton et al., 1960; Wenkert et al., 1959), due to isomerization at the spiro center through Mannich/retro-Mannich reactions (Figure 1). The composition of the mixture depends on temperature, pH, and solvent polarity and complicates the study of the biological profiles of single isomers (Laus, 1998; Laus et al., 1996; Kang et al., 2004; Yuan et al., 2008). The synthesis of natural products containing the secoyohimbane core has been a subject of significant recent interest (Deiters et al., 2006; Ito et al., 2001; Martin and Mortimore, 1990; Stahl et al., 1996; Lerchner and Carreira, 2002, 2006). Inspired by the biological importance of this scaffold, the instability of its isomers, and the lack of efficient synthesis methods for the generation of secoyohimbane-inspired compound collections, we sought to develop an asymmetric synthesis approach to access nonisomerizable secoyohimbanes. We report herein a highly enantioselective synthesis method that gives access to the secoyohimbane scaffold. Our approach employs asymmetric iminium organocatalysis (Marqués-López et al., 2010; Grondal et al., 2010; Bertelsen and Jørgensen, 2009; Melchiorre et al., 2008; Enders et al., 2007) as key stereodirecting method, is highly practical, uses

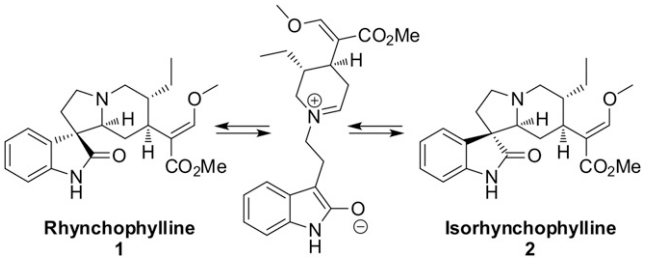


Figure 1. Isomerization of Secoyohimbane Alkaloids
In nature, rhynchophylline and isorhynchophylline occur as pairs of interconvertible isomers. The isomerization proceeds via a retro-Mannich reaction involving an open-ring intermediate followed by Mannich reaction.

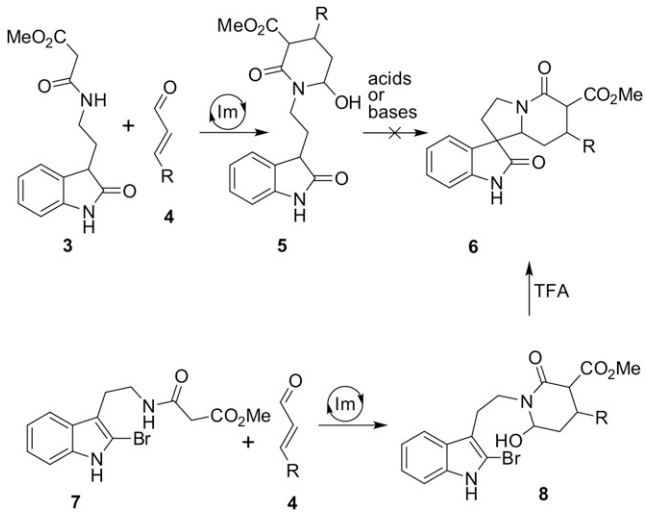


Figure 2. Planned Synthesis Route to the Secoyohimbane Scaffold
The initial approach based on the use of intermediate 3 for the synthesis of spirocyclic products was not successful. Desired secoyohimbane scaffold was obtained using intermediate 7 in key organocatalytic reaction cascade in the second approach. Im, iminium catalysis.

readily accessible starting materials, and generates the tetracyclic core, incorporating one all-carbon quaternary spirocenter (Bella and Gasperi, 2009; Zhou et al., 2010; Trost and Brennan, 2009; Steven and Overman 2007; Trost and Jiang 2006) and three tertiary centers in a multistep, one-pot transformation. While we were exploring this strategy, Wu et al. (2012) and Zhang et al. (2013) independently investigated a very similar approach. Motivated by the reasoning of biology-oriented synthesis (Wetzel et al., 2011; Dückert et al., 2012; Wilk et al., 2010; Kaiser et al., 2008; Lachance et al., 2012; Over et al., 2013) as approach for the design and synthesis of natural product-inspired compound collections enriched by molecules with diverse bioactivity, we investigated the synthesized compounds in cell-based assays monitoring neurite outgrowth from primary hippocampal neurons and embryonal stem cell (ESC)-derived motor neurons. The screen identified library members with a pronounced influence on the complexity of neuronal network formation and polarized cell growth.

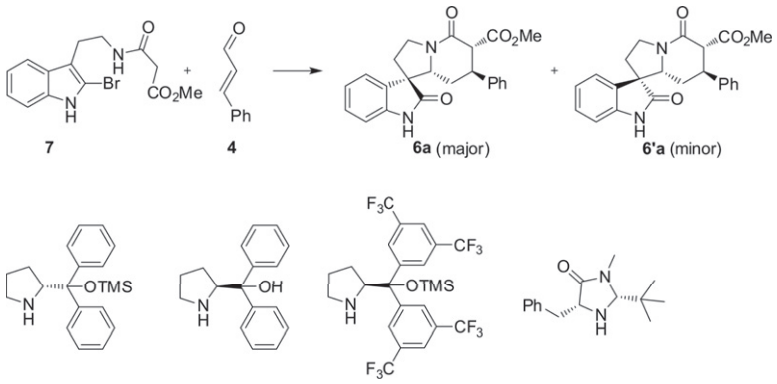
RESULTS AND DISCUSSION

Development of an Enantioselective Catalyzed Synthesis of a Secoyohimbane Library

Initially, we focused on the development of an asymmetric catalytic domino Michael-Mannich reaction of racemic oxindole derivative (3) and α,β-unsaturated aldehydes (4) to yield a secoyohimbane (6) (Figure 2). The products (6) would contain an amide function instead of the tertiary amine of the indolizine, which should prevent isomerization via Mannich/retro-Mannich reactions. However, despite extensive examination of various reaction conditions, the formation of the target secoyohimbanes (6) was not observed and only cyclic hemiaminals (5) were obtained as stable intermediates. All attempts to convert intermediates (5) to products (6) via formation of an acyliminium ion and Mannich reaction in the presence of various Brønsted acids and bases failed. In the light of this failure, a new approach for the preparation of the secoyohimbane scaffold needed to be developed. We envisioned to use 2-bromoindole derivative (7) (Figure 2; Miyamoto et al., 2006; Miyake et al., 2004) as decisive starting material, in which the 2-bromo substituent would serve a double function. On the one hand, it was meant to serve as a protecting group, preventing cyclization to the two position of the indole in 8 (masked by the bromine), and due to that, the formation of the tetrahydro-β-carboline cannot occur, redirecting the attack of the electrophile to the three position. Thus, we assumed that the catalytic enantio- and regioselective addition of 7 to α,β-unsaturated aldehydes would result in the formation of hemiaminal (8) (Brandau et al., 2006; Marigo et al., 2006; Franzén and Fisher, 2009; Hayashi et al., 2009; Valero et al., 2009; Zhang and Franzén, 2010). On the other hand, the bromine served the purpose to enable the diastereoselective conversion of the intermediate hemiaminal (8) (after treatment with trifluoroacetic acid (TFA) performing a Friedel-Crafts addition followed by hydrolysis) into the target secoyohimbane scaffold (6).

Gratifyingly, orientating experiments employing 2-bromoindole (7) and cinnamic aldehyde in the presence of an organocatalyst indicated that the strategy was indeed viable and that the spiroindolinones were formed.

In order to establish favorable reaction conditions, under which the desired secoyohimbane-inspired compounds would be obtained in preparatively viable yields and with high stereoselectivity, as model reaction, the addition of nucleophile (7) to cinnamic aldehyde in the presence of different organocatalysts (A–D) in several solvents and in the presence of potassium acetate as additive was optimized. The search for a suitable solvent (Figure 3, entries 1–10) revealed that the best results for 6a with respect to enantiomeric excess, diastereoselectivity, and yield were obtained in methanol with organocatalyst (A) (Figure 3, entry 3). We note that the presence of the additive is essential for product formation in all solvents tested. Diphenylprolinol trimethylsilylether (A) gave a high level of enantiocontrol, whereas diphenylprolinol (B) itself did not catalyze the reaction (Figure 3, entry 8). Jorgensen's catalyst (C) or MacMillan's imidazolidinone (D) led to lower levels of enantioselectivity (Figure 3, entries 9 and 10). To further optimize the reaction conditions, we varied the additives (Figure 3, entries 11–16). We found that the addition of caesium acetate slightly improved the yield of product 6a and the enantioselectivity (Figure 3, entry 17). Further



Entry[a]	Catalyst	mol%	Additive	mol%	Solvent	Time [hr]	Yield [%][b]	dr[c]	ee [%][d]
1	**A**	20	KOAc	120	CH_2Cl_2	48	-	-	-
2	**A**	20	KOAc	120	PhMe	48	-	-	-
3	**A**	20	KOAc	120	MeOH	16	51	3/1	91/90
4	**A**	20	KOAc	120	EtOH	16	34	1/1	90/90
5	**A**	20	KOAc	120	i-PrOH	16	49	2/1	89/87
6	**A**	20	KOAc	120	TFE	16	52	1/1	86/87
7	**A**	20	KOAc	120	MeCN	16	52	2/1	92/91
8	**B**	20	KOAc	120	MeOH	48	-	-	-
9	**C**	20	KOAc	120	MeOH	16	48	1/1	85/84[e]
10	**D**	20	KOAc	120	MeOH	16	41	4/1	58/53
11	**A**	20	AcOH	120	MeOH	16	48	2/1	91/87
12	**A**	20	LiOAc	120	MeOH	16	50	2/1	90/87
13	**A**	20	NaOAc	120	MeOH	16	44	3/1	90/87
14	**A**	20	RbOAc	120	MeOH	16	55	3/1	91/90
15	**A**	20	CsOAc	120	MeOH	16	57	3/1	92/90
16	**A**	20	BzOH	120	MeOH	26	41	3/1	92/91
17	**A**	10	CsOAc	120	MeOH	16	56	3/1	92/90
18	**A**	10	CsOAc	60	MeOH	16	58	3/1	92/90
19	**A**	10	CsOAc	20	MeOH	16	45	3/1	91/90
20[f]	**A**	10	CsOAc	60	MeOH	20	63	5/1	95/91

Figure 3. Exploration of Different Reaction Conditions

[a]Reaction conditions: aldehyde (4) (1.0 equiv), indole (7) (1.2 equiv), additive, and catalyst were stirred in solvent at ambient temperature for the given time. Trifluoroacetic acid was added at −60°C, and the reaction mixture was warmed up to room temperature.

[b]Isolated yields of 6a after column chromatography.

[c]Determined by 1H nuclear magnetic resonance (NMR) analysis.

[d]Determined by high-performance liquid chromatography (HPLC) analysis. The ee values for both spiroindolinones 6a and 6′a.

[e]Opposite enantiomer.

[f]At 0°C.

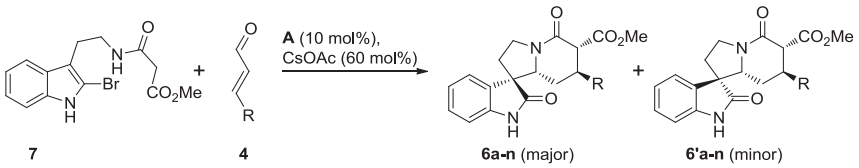
TFE, 2,2,2-trifluorethanol.

experiments revealed that the catalyst loading could be reduced to 10 mol % and that substoichiometric amounts of caesium acetate suffice (Figure 3, entries 18 and 19). The enantioselectivity could be increased from 92% to 95% by lowering the reaction temperature to 0°C (Figure 3, entry 20). Further decrease of the reaction temperature led to low conversion rates and prolonged reaction times.

Notably, an interconversion of single isolated isomers was not observed upon storage under the reaction conditions for 4 weeks at ambient temperature.

With the optimized reaction conditions identified, we investigated the substrate scope with regard to various α,β-unsaturated aldehydes (Figure 4). A broad range of aldehydes with electron-donating and -withdrawing substituents, para-, meta-, and orthosubstituted cinnamaldehydes as well as acroleines substituted with a heterocycle in the β-position can be employed successfully in the organocatalyzed enantioselective cascade reaction. The secoyohimbane-inspired products were obtained in preparatively viable yields and with excellent enantioselectivity (88%–96% enantiomeric excess [ee]). The absolute configuration of the predominantly formed stereoisomer was determined unambiguously by means of single-crystal X-ray analysis of compound 6e (Figure S1 available online).

Mechanistically, the first step of the developed transformation is a stereo- and regioselective Michael addition of the 2-bromoindole (7) to the α,β-unsaturated aldehyde, which establishes two stereocenters (Figure 5). Although 7 contains several nitrogen- and two carbon-nucleophiles, only addition of the acidic methylene group to the unsaturated aldehyde was observed, which leads to intermediate (9). The stereochemical course of the organocatalyzed conjugate addition is in accordance with relevant results reported for the addition of malonic acid derivatives to enals (Brandau et al., 2006; Marigo et al., 2006; Franzén and Fisher, 2009; Hayashi et al., 2009; Valero et al., 2009; Zhang and Franzén, 2010). Intermediate (9) then undergoes spontaneous cyclization to form hemiaminal (8). Elimination of water after protonation by the Brønsted acid leads to formation of iminium intermediate (11), which is in equilibrium with enamide (10). The iminium intermediate (11) undergoes an electrophilic Friedel-Crafts cyclization to yield spirocycle (12), embodying an imidoyl bromide, which is hydrolyzed in the presence of Brønsted acid and water to yield target spiroindolinone (6). Interestingly, Mannich cyclization of cyclic hemiaminal (5), which could be obtained from 8 by hydrolysis of the imidoyl bromide does not occur. In order to rationalize the stereochemical preference in the spirocyclization, we assume that two competing transition states TS1 and TS2 may be passed, in which the indole attacks the acyl iminium intermediate anti to substituent R (Figure S2). In TS1, which leads to the observed configuration of the products, the bromine points away from the ester substituent. Rotation around the bond connecting C3 of the indole with the iminium-N would lead to TS2 and yield the isomeric spiro configuration. TS2 is less preferred than TS1, due to unfavorable interactions between the bromine and the ester group.



Entry[a]		R	Time [hr]		Yield [%][b]	dr[c]	ee [%][d]
1	**4a**		20	**6a**	63	5/1	95
2	**4b**	CF_3	20	**6b**	51	5/1	94
3	**4c**	NO_2	16	**6c**	55	2/1	92
4	**4d**	F	16	**6d**	25	2/1	95
5	**4e**	Br	16	**6e**	52	10/1	95
6	**4f**	OMe	40	**6f**	55	10/1	92
7	**4g**	Me	20	**6g**	69	5/1	96
8	**4h**		40	**6h**	60	5/1	89
9	**4i**		36	**6i**	62	10/1	95
10	**4j**		40	**6j**	61	15/1	95
11	**4k**	F, Cl	20	**6k**	35	1.2/1	96
12	**4l**	Cl, Cl	20	**6l**	46	1.5/1	90
13	**4m**		30	**6m**	58	4/1	88
14	**4n**		40	**6n**	42	8/1	93

Figure 4. Results of the Enantioselective Catalyzed Synthesis of Secoyohimbane-Inspired Spirocyclic Indolinones

[a]For detailed experimental procedures and characterization of the products, see the Supplemental Information.

[b]Isolated yields of the single diastereomer after column chromatography.

[c]Determined by ^{1}H NMR analysis.

[d]Determined by HPLC analysis for the major isomer.

See also Figure S1 (entry 5, compound 6e).

Biological Validation of the Secoyohimbane-Inspired Library

In light of the use of *Uncaria* species for the treatment of neurodegenerative diseases, we investigated whether the natural product-inspired spirocyclic indolinones positively influence neurite outgrowth from primary rat hippocampal neurons (Dotti et al., 1988). In an initial screen, the major diastereomers (6a–6n) were dissolved in DMSO and screened at a final concentration of 10 μM for their influence on quantitative membrane outgrowth by monitoring incorporation of a fluorescent dye into the increasing amount of cell membrane formed. Fluorescence intensity is higher with increasing growth and, thereby, formation of neuronal membranes. Analysis of the fluorescent staining revealed that 2-naphthyl derivative (6i) and 3-furyl derivative (6n) were most active and exposure of the neuronal cells to these compounds led to dramatic outgrowth of the neuronal membrane. Therefore, they were chosen for more detailed characterization.

In the subsequent in-depth analysis, changes in the morphology of neurons exposed to the 2-naphtyl (6i) and 3-furyl (6n) derivatives were investigated. Retinoic acid (RA) and brain-derived neurotrophic factor (BDNF) were used as positive controls. RA and BDNF bind to their corresponding receptors, promoting polymerization of the cytoskeleton preceding the establishment of a more developed complex neuronal network (Theus et al., 2006; Clagett-Dame et al., 2006; Yasuda et al., 2007; Kozisek et al., 2008). Furthermore, the natural products rhynchophylline (Rhy) and isorhynchophylline (IsoRhy) were included in the experiment. Rhynchophylline acts as a noncompetitive antagonist of the *N*-methyl-D-aspartate receptor and influences the modulation of calcium and potassium ion channels (Kang et al., 2004; Xu et al., 2012). However, the exact mechanisms of rhynchophylline activity and its targets are not known. The confocal images acquired after exposure of the primary cultured rat hippocampal neurons to small molecules and the observed morphological changes were quantitatively analyzed (Figure 6; see also the Supplemental Information).

Exposure of primary cultured rat hippocampal neurons to rhynchophylline shows a strong effect on the development of neurons. Rhynchophylline increased the complexity of the neuronal network formed, since the total neurite length and single neurite length are higher in comparison to control cells (Figures 6I and 6K). However, in the presence of the natural product, the number of neurites remains comparable to treatment with DMSO (Figure 6J). The formed neurites are longer in comparison to treatment with the controls, BDNF and RA (Figure 6J). Furthermore, the length of the axon is similar to the value recorded for application of the positive control (RA effect) (Figure 6H). This observation suggests that

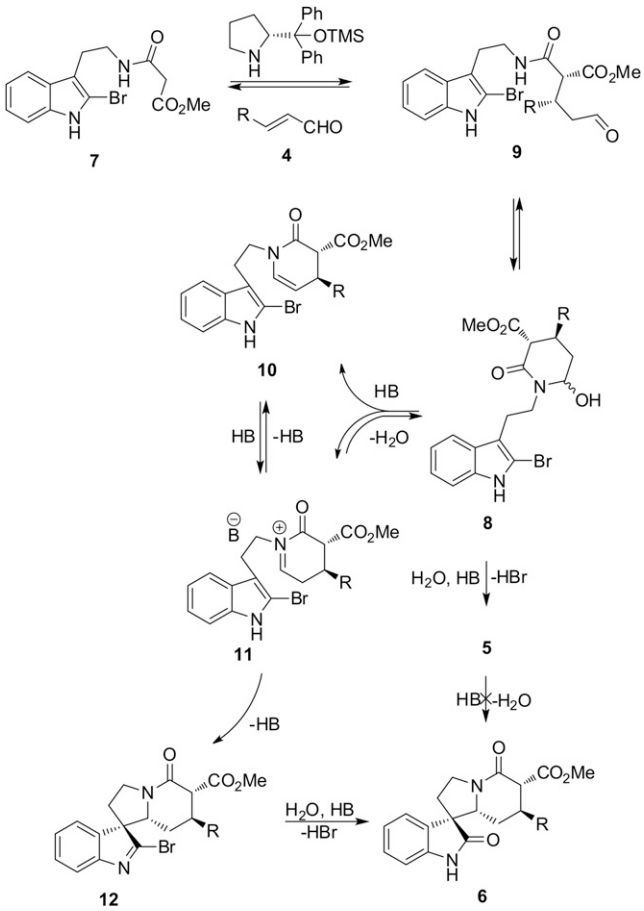


Figure 5. Proposed Mechanism of Organocatalytic Synthesis of the Secoyohimbane Scaffold

The mechanism of spirocyclic products (6) formation is included in the sequence of reactions. The key step of the reactions sequence is enantioselective organocatalytic Michael addition to unsaturated aldehydes to form an intermediate (9). Following cyclization of 9 generates N-acyl-iminium ion (11), which under the reaction conditions undergoes Mannich reaction to give the desired products (6). HB, Brønsted acid; HBr, hydrobromic acid.
See also Figure S2.

rhynchophylline induces cytoskeletal rearrangement toward formation of more polarized cells undergoing maturation. The results support the known neuroprotective activity of rhynchophylline (Shimada et al., 1999). Interestingly, the spiroisomeric isorhynchophylline unfavorably impairs the development of primary cultured rat hippocampal neurons (Figure 6). To our delight, 3-furyl derivative (6n), which differs in substituent pattern and absolute configuration at $C8_{a'}$ from rhynchophylline, yet clearly has a structure resembling the natural product and displays an activity profile very similar to the natural alkaloid (Figure 6). Replacement of the 3-furyl group (6n) with the 2-naphtyl group (6i) did not result in a difference in outgrowth of the neuronal membrane. However, administration of derivative 6i led to the formation of complex neuronal networks, which is reflected in a substantial increase of single neurite length and lower number of neurites per neuron. This finding may reflect a different mode of action of compounds 6i and 6n, rhynchophylline. In general, the morphological changes observed for the primary hippocampal cells after exposure to the spirocyclic natural product-inspired secoyohimbanes indicate that their mode of action may include modulation of cytoskeletal rearrangement processes and proteins involved therein and possibly an influence on growth cone development (Dent and Gertler, 2003).

The results obtained with rat primary hippocampal neurons were also validated using a different approach, employing mouse motor neurons derived from ESCs to determine the total neurite length, the number of branch points and the maximal length of neurites. Trophic factors, such as BDNF, neurotrophin-4, ciliary neurotrophic factor, and glial cell line-derived neurotrophic factor (GDNF), have different effects on neurons in vitro or in vivo, including axonal growth, neurogenesis, and an increase in survival (Airaksinen and Saarma, 2002; Bothwell, 1995; Zurn et al., 1996), and, as such, have been used as therapeutic agents in a wide range of human diseases (Yamamoto et al., 2001; Schindowski et al., 2008; Henriques et al., 2010; Masi and Brovedani, 2011). However, with age and in pathological conditions, the production of neurotrophic factors from satellite cell or the target organs can be reduced and lead to increased susceptibility to nerve degeneration and poor neuronal survival. For that reason, in all of the experiments, DMSO was considered as a negative control and GDNF as a positive control to compare with the activity of the molecules tested, since this factor shows a very potent effect on motor neurons among these neurotrophic factors. The images acquired after exposure of ESC-derived motor neurons to small molecules and the observed morphological changes were quantitatively analyzed with Cellomics Bioapplication Neurite Profiling (Figure 7).

In the experiments, *p*-trifluoromethylphenyl derivative (6b), *p*-methoxyphenyl derivative (6f), and 3-furyl derivative (6n), were identified as hits. In accordance to the neurotrophic hypothesis (Oppenheim, 1989), axonal branches from various neurons compete for neurotrophic molecules, and the access to neurotrophic molecules from the target tissue regulates the number of neurons: the survival of cells with a neurotrophic deficit is compromised, while those with a correct trophic support can survive. Small changes in the neurotrophic support can cause changes in the number of motor neurons obtained in different wells, with big effect on the neurite branching and length, especially after 7 days in vitro. Because of that, the values of the error bars in the graphs are disproportionately large. Thus, neurite arborization and neuronal survival can be used as an indicator of the neurotrophic activity of different molecules. According to the results in the graphs, compound *p*-trifluoromethylphenyl derivative (6b) is the most active in all experiments composed of neurite total length, number of branch points, and neurite maximal length. The most remarkable effect is shown in neurite maximal length, which means that the cells treated with this molecule have the longest neurites. Compound 6f is less active, but still presents the same type of activity in all experiments. In general, the activity of these spirocyclic indolinones are below the values of the powerful trophic factor GDNF; however, they provide good starting points for further structural modifications aimed at increasing their neurotrophic activity.

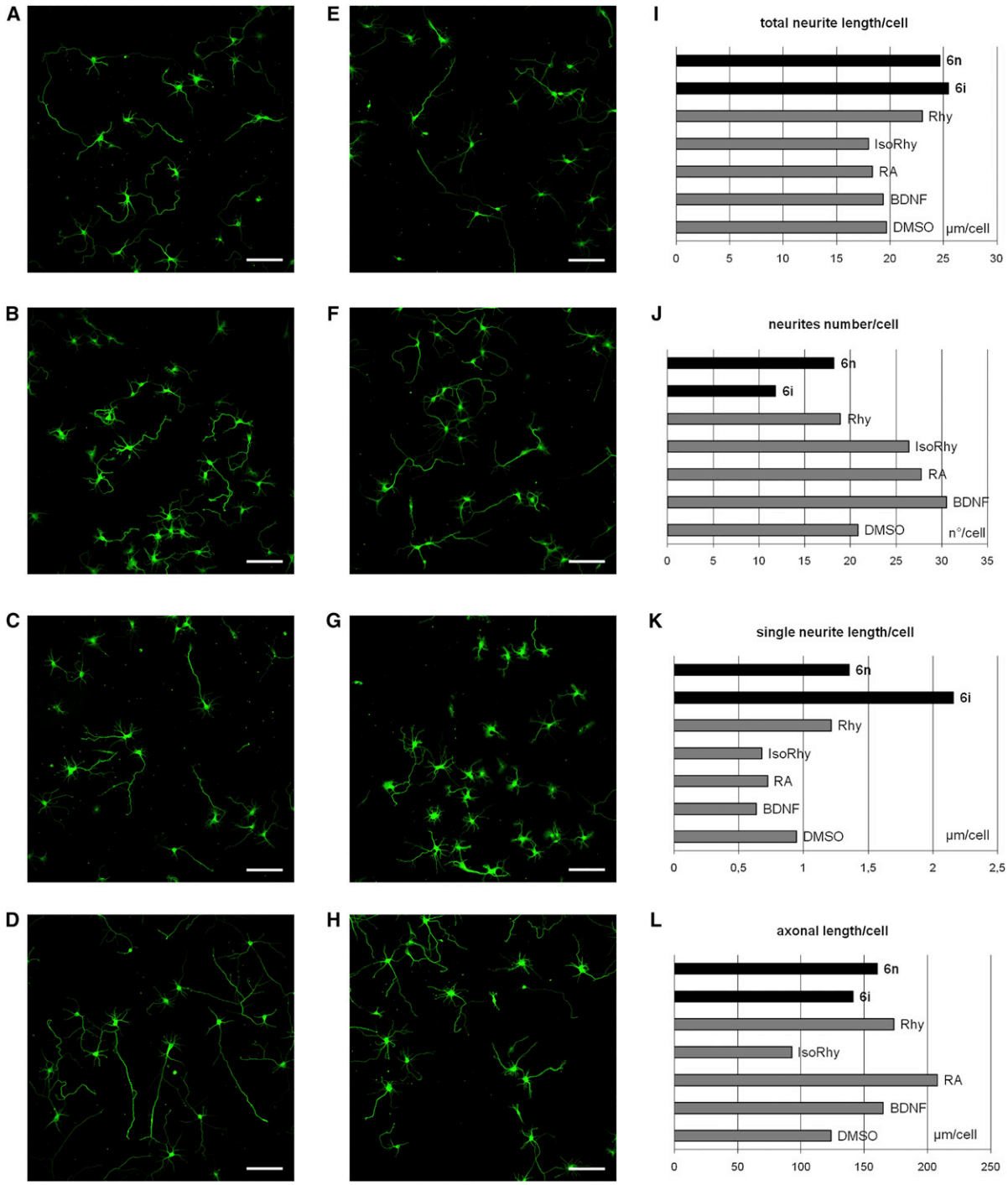


Figure 6. Neurite Outgrowth-Promoting Activity on Primary Cultured Rat Hippocampal Neurons and Quantification of Morphological Changes

(A) Morphology of neurons of untreated cells.
(B) Morphology of neurons in the presence of DMSO.
(C) Morphology of neurons treated with BDNF.
(D) Morphology of neurons treated with retinoic acid.
(E) Morphology of neurons treated with 10 μM of 6n.
(F) Morphology of neurons treated with 10 μM of 6i.
(G) Morphology of neurons treated with 10 μM of isorhynchophylline.
(H) Morphology of neurons treated with 10 μM of rhynchophylline.
(I) Total neurite length per cell.

(legend continued on next page)

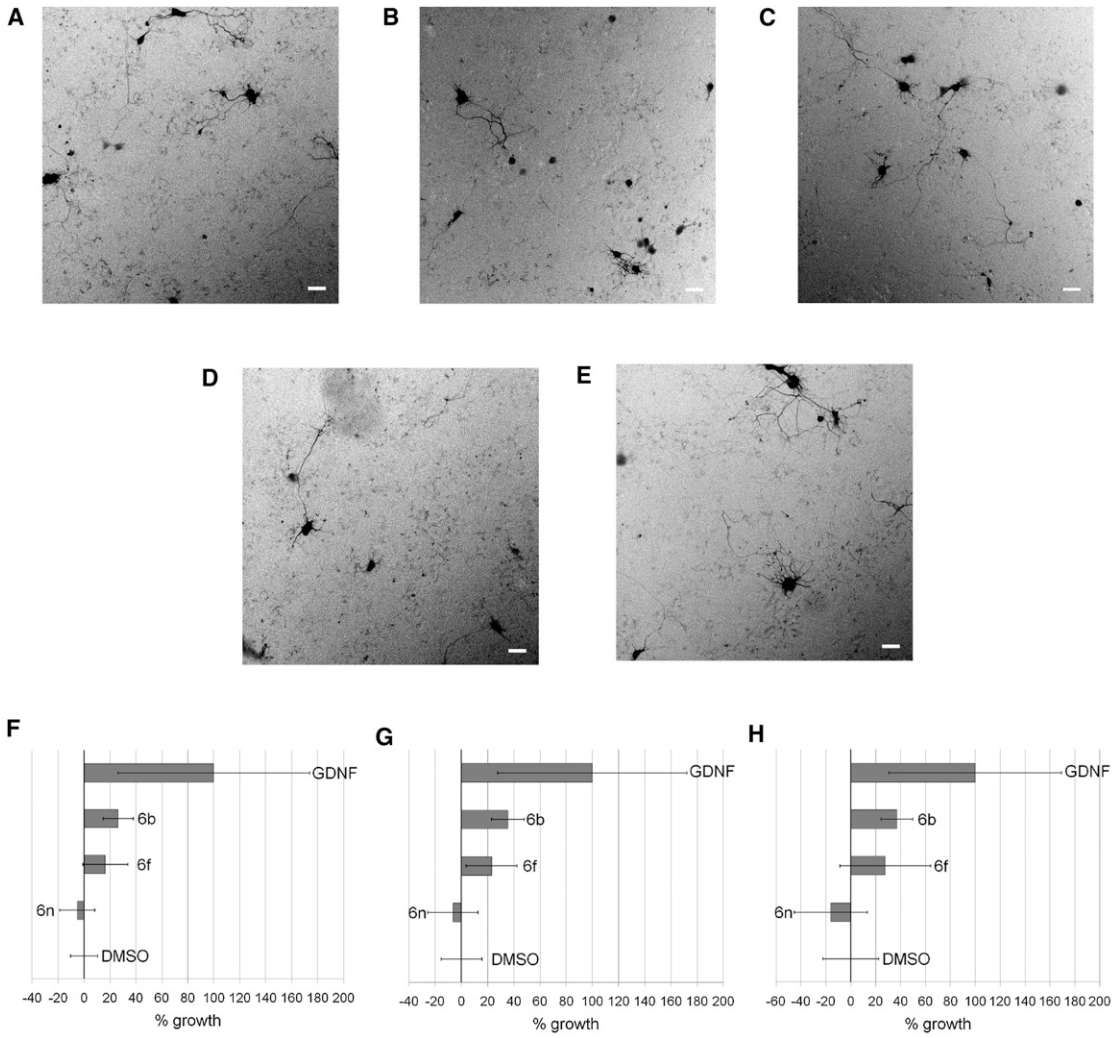


Figure 7. Neurite Outgrowth-Promoting Activity and Quantification of Morphological Changes on ESC-Derived Motor Neurons

(A) Morphology of neurons treated with 10 μM of 6b.
(B) Morphology of neurons treated with 10 μM of 6f.
(C) Morphology of neurons treated with 10 μM of 6n.
(D) Morphology of neurons in the presence of DMSO.
(E) Morphology of neurons treated with GDNF.
(F) Neurite total length.
(G) Branch points.
(H) Maximum of neurite length.
Scale bar: 40 μm. Error bars show SD (n = 4).

In conclusion, we developed a highly practical catalytic method for the synthesis of a collection of complex natural product-inspired compounds with the basic scaffold of the secoyohimbane alkaloids. The polycyclic heterocycles embodying one quaternary and three tertiary stereocenters are obtained with excellent enantioselectivity and with preparatively viable yields from simple and readily available starting materials in a one-pot multistep reaction sequence. Investigation of this family of spirocyclic indolinones for neurite outgrowth-promoting activity employing two different neuronal cell lines revealed remarkable neurotrophic activity and provided insight for further structural modification to arrive at more potent compounds.

(J) Number of neurites per cell.
(K) Length of single neurite per cell.
(L) Axonal length per cell.
Scale bar: 100 μm. CTRL, untreated cells.
See also Figure S3.

SIGNIFICANCE

Many neurological diseases are characterized by a progressive loss of neuronal activity, and novel approaches aimed at identification of small molecules with neurotrophic properties are of major importance. Natural products with neurotrophic activity provide evolutionary validated core structures for the synthesis of compound classes endowed with neurite growth-promoting activity. The development of synthesis methods that make such compound classes accessible in a very practical, flexible, and enantioselective manner has high significance. Therefore, a highly efficient diastereo- and enantioselective and organocatalyzed cascade access to the secoyohimbane scaffold of the neuromodulatory alkaloid rhynchophylline was developed that generates four stereocenters in a one-pot multistep reaction sequence. Investigation of a compound collection synthesized by means of this method revealed a class of neurotrophic compounds.

EXPERIMENTAL PROCEDURES

All products were fully characterized. Spectral data for all compounds are provided in the Supplemental Information.

Typical Procedure for the Organocatalyzed Synthesis

To a solution of the aldehyde (4) (0.4 mmol) and the catalyst (10 mol %, 40 μmol) in MeOH (0.5 ml) were added CsOAc (60 mol %, 0.24 mmol) and 3-(2-(1H-indol-3-yl)ethylamino)-3-oxopropanoate (7) (1.2 eq., 0.48 mmol) in MeOH (0.5 ml) at 0°C. The reaction mixture was stirred at 0°C for 20–40 hr, cooled to −80°C, and TFA (10 eq) was added. The reaction mixture was warmed to room temperature, passed through a short plug of silica, and concentrated under reduced pressure. Purification by column chromatography on silica gel (ethyl acetate/dichloromethane = 1/5 → 1/2) yielded the product.

ACCESSION NUMBERS

The Cambridge Crystallographic Data Centre accession number for the compound 6e reported in this paper is CCDC 915444.

SUPPLEMENTAL INFORMATION

Supplemental Information includes three figures and Supplemental Experimental Procedures and can be found with this article online at http://dx.doi.org/10.1016/j.chembiol.2013.03.011.

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